

Amendments to the claims

1. to 6. (Cancelled)

7. (Currently amended) A process for generating and detecting recombinant DNA sequences in *Saccharomyces cerevisiae* comprising the steps of: ~~Process according to according to any one of claims 1 to 3, wherein the first diploid cell is generated by fusing a haploid *S. cerevisiae* cell bearing in a locus of its genome the first recombination cassette with a haploid *S. cerevisiae* cell bearing in an allelic position the second recombination cassette.~~

a) inserting a first DNA sequence to be recombined between first and second marker sequences located adjacently on a first cloning vehicle, wherein the first cloning vehicle is plasmid pMXY9 (deposited as DSM 17010);

b) excising from the first cloning vehicles obtained in a) a first fragments bearing the first recombination cassette, whereby the first recombination cassette comprises the first DNA sequence flanked by the first and second marker sequences, and the first recombination cassette in turn is flanked by targeting sequences,

c) inserting a second DNA sequence to be recombined between third and fourth marker sequences located adjacently on a second cloning vehicle, wherein the second cloning vehicle is plasmid pMXY12 (deposited as DSM 17011);

d) excising from the second cloning vehicles obtained in c) a second fragments bearing the second recombination cassette, whereby the second recombination cassette comprises the second DNA sequence flanked by the third and fourth marker sequences, and the second recombination cassette in turn is flanked by targeting sequences,

e) transforming the first and second fragments bearing the recombination cassettes with flanking targeting sequences obtained in b) and d) separately into *S. cerevisiae* haploid cells, whereby the targeting sequences direct the integration of the cassettes into that locus to which they are homologous, in order to obtain a first haploid cell comprising the first cassette and a second haploid cell comprising the second cassette;

f) fusing the first haploid cell bearing in a locus of its genome the first recombination cassette with the second haploid cell comprising in an allelic position the second recombination cassette in order to produce a heterozygous diploid cell;

g) inducing the sporulation of the heterozygous diploid cell obtained in f) and

h) isolating haploid cells containing recombination cassettes in which first recombined DNA sequences are flanked by at least the first and fourth marker sequences, and haploid cells containing recombination cassettes in which second recombined DNA sequences are flanked by at least the second and the third marker sequences.

8. (Currently amended) A process for generating and detecting recombinant DNA sequences in *Saccharomyces cerevisiae* comprising the steps of: ~~Process according to according to any one of claims 1 to 3, wherein the first diploid cell is generated by mating a haploid *S. cerevisiae* cell bearing in a locus of its genome the first recombination cassette with a haploid *S. cerevisiae* cell bearing in an allelic position the second recombination cassette.~~

a) inserting a first DNA sequence to be recombined between first and second marker sequences located adjacently on a first cloning vehicle, wherein the first cloning vehicle is plasmid pMXY9 (deposited as DSM 17010);

b) excising from the first cloning vehicles obtained in a) a first fragments bearing the first recombination cassette, whereby the first recombination cassette comprises the first DNA sequence flanked by the first and second marker sequences, and the first recombination cassette in turn is flanked by targeting sequences,

c) inserting a second DNA sequence to be recombined between third and fourth marker sequences located adjacently on a second cloning vehicle, wherein the second cloning vehicle is plasmid pMXY12 (deposited as DSM 17011);

d) excising from the second cloning vehicles obtained in c) a second fragments bearing the second recombination cassette, whereby the second recombination cassette comprises the second DNA sequence flanked by the third and fourth marker sequences, and the second recombination cassette in turn is flanked by targeting sequences,

e) transforming the first and second fragments bearing the recombination cassettes with flanking targeting sequences obtained in b) and d) separately into *S. cerevisiae* haploid cells, whereby the targeting sequences direct the integration of the cassettes into that locus to which they are homologous, in order to obtain a first haploid cell comprising the first cassette and a second haploid cell comprising the second cassette;

f) mating the first haploid cell bearing in a locus of its genome the first recombination cassette with the second haploid cell comprising in an allelic position the second recombination cassette in order to produce a heterozygous diploid cell;

g) inducing the sporulation of the heterozygous diploid cell obtained in f) and

h) isolating haploid cells containing recombination cassettes in which first recombined DNA sequences are flanked by at least the first and fourth marker sequences, and haploid cells containing recombination cassettes in which second recombined DNA sequences are flanked by at least the second and the third marker sequences.

9. (Currently amended) A process for generating and detecting recombinant DNA sequences in *Saccharomyces cerevisiae* comprising the steps of:

~~Process according to claim 7 or 8, wherein haploid cells bearing the first or second recombination cassette are generated by:~~

~~a) inserting a the first DNA sequence to be recombined between the first and the second marker sequences located adjacently on a first cloning vehicle, wherein the first cloning vehicle is plasmid pMXY9 (deposited as DSM 17010); and inserting the second DNA sequence to be recombined between the third and the fourth marker sequences located adjacently on a second cloning vehicle, whereby the respective two marker sequences are flanked by targeting sequences which are homologous to a defined locus of the *S. cerevisiae* genome;~~

~~b) excising from the first cloning vehicles obtained in a) a first fragments bearing the first recombination cassette and the second recombination cassette, respectively, whereby the first recombination cassette each of the cassettes comprises the first DNA sequence to be~~

~~recombined~~ flanked by the first and second ~~respective two~~ marker sequences, and the
~~each~~ first recombination cassette in turn is flanked by targeting sequences,

c) inserting a second DNA sequence to be recombined between third and fourth marker
sequences located adjacently on a second cloning vehicle, wherein the second cloning
vehicle is plasmid pMXY12 (deposited as DSM 17011);

d) excising from the second cloning vehicles obtained in c) a second fragments bearing
the second recombination cassette, whereby the second recombination cassette comprises
the second DNA sequence flanked by the third and fourth marker sequences, and the
second recombination cassette in turn is flanked by targeting sequences,

ee) transforming the first and second fragments bearing the recombination cassettes with
flanking targeting sequences obtained in b) and d) ~~separately~~ into *S. cerevisiae* diploid
cells, whereby the targeting sequences direct the integration of the cassettes into that
locus to which they are homologous, in order to obtain diploid cells heterozygous for the
first cassette, or the second cassette,

~~fd)~~ inducing ~~separately~~ the sporulation of the heterozygous diploid cells obtained in c)
and

ge) isolating haploid cells containing the first cassette and expressing the first and fourth
~~second~~ marker sequences and separately haploid cells containing the second cassette and
expressing the second ~~third~~ and the third ~~fourth~~ marker sequences.

10. (Cancelled)

11. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the
diploid *S. cerevisiae* cells used for transformation are auxotrophic for at least two
nutritional factors.

12. (Currently amended) The process ~~Process~~ according to claim 11, wherein the
diploid cells are homozygous for the *ura3-1* allele and the *trp1-1* allele, which render them
auxotrophic for uracil and tryptophan, respectively.

13. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the

diploid cells used for transformation are resistant to at least two antibiotics.

14. (Currently amended) The process ~~Process~~ according to claim 13, wherein the diploid cells are homozygous for the can1-100 allele and the cyh2R allele, which render them resistant to canavanine and cycloheximide, respectively.

15. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein diploid cells of the *S. cerevisiae* strain MXY47 are used for transformation, which are homozygous for the alleles ura3-1, trp1-1, can1-100 and cyh2R and heterozygous for the msh2::KanMX mutation.

16. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the *S. cerevisiae* cells have a functional mismatch repair system.

17. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the *S. cerevisiae* cells are transiently or permanently deficient in the mismatch repair system.

18. (Currently amended) Process according to claim 17, wherein the transient or permanent deficiency of the mismatch repair system is due to an mutation and/or an inducible expression or repression of one or more genes involved in the mismatch repair system, a treatment with an agent that saturates the mismatch repair system and or a treatment with an agent that globally impairs the mismatch repair.

19. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the first and the second recombination cassettes are integrated in the BUD31-HCM1 locus on chromosome III of the *S. cerevisiae* genome.

20. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the first and the second DNA sequences to be recombined diverge by at least 1 nucleotide.

21. (Currently amended) The process ~~Process~~ according to claim 9 4, wherein the

first and the second DNA sequences to be recombined are derived from organisms other than and including *S. cerevisiae*.

22. (Currently amended) The process ~~Process~~ according to claim 9 ~~4~~, wherein the first and the second DNA sequences to be recombined comprise one or more non-coding sequences and/or one or more protein-coding sequences.

23. (Currently amended) The process ~~Process~~ according to claim 9 ~~4~~, wherein the marker sequences are selected from the group consisting of nutritional markers, pigment markers, antibiotic resistance markers, antibiotic sensitivity markers, primer recognition sites, intron/exon boundaries, sequences encoding a particular subunit of an enzyme, promoter sequences, downstream regulated gene sequences and restriction enzyme sites.

24. (Currently amended) The process ~~Process~~ according to claim 23, wherein the first and third marker sequences are nutritional markers, the gene products of which can compensate an auxotrophy of a *S. cerevisiae* cell.

25. (Currently amended) The process ~~Process~~ according to claim 24, wherein the first marker sequence is URA3, the gene product of which can confer uracil prototrophy to an uracil auxotrophic *S. cerevisiae* cell.

26. (Currently amended) The process ~~Process~~ according to claim 24, wherein the third marker sequence is TRP1, the gene product of which can confer tryptophan prototrophy to an tryptophan auxotrophic *S. cerevisiae* cell.

27. (Currently amended) The process ~~Process~~ according to claim 23, wherein the second and fourth marker sequences are antibiotic sensitivity markers, the gene products of which can confer sensitivity to an antibiotic to a *S. cerevisiae* cell which is resistant to that antibiotic.

28. (Currently amended) The process ~~Process~~ according to claim 27, wherein the

second marker sequence is CAN1, the gene product of which can confer sensitivity to canavanine to a canavanine-resistant *S. cerevisiae* cell.

29. (Currently amended) The process ~~Process~~ according to claim 27, wherein the fourth marker sequence is CYH2, the gene product of which can confer sensitivity to cycloheximide to a cycloheximide-resistant *S. cerevisiae* cell.

30. (Currently amended) The process ~~Process~~ according to claim 9 ~~4~~, wherein haploid cells containing recombination cassettes with either first, second, third or fourth recombined DNA sequences are identified by PCR processes in order to detect the presence of the respective marker combination.

31. (Currently amended) The process ~~Process~~ according to claim 9 ~~4~~, wherein haploid cells containing recombination cassettes with either first, second, third or fourth recombined DNA sequences are identified by plating the haploid cells on media that select for the molecular linkage on the same DNA molecule of the respective marker combination.

32. (Currently amended) The process ~~Process~~ according to claim 31, wherein haploid cells containing first recombined DNA sequences are plated on a medium that selects for molecular linkage on the same DNA molecule of the first and the fourth marker sequences.

33. (Currently amended) The process ~~Process~~ according to claim 31, wherein haploid cells containing second recombined DNA sequences are plated on a medium that selects for molecular linkage on the same DNA molecule of the second and the third marker sequences.

34. (Currently amended) The process ~~Process~~ according to claim 31, wherein haploid cells containing third recombined DNA sequences are plated on a medium that selects for molecular linkage on the same DNA molecule of the first and the second

marker sequences.

35. (Currently amended) The process ~~Process~~ according to claim 31, wherein haploid cells containing fourth recombined DNA sequences are plated on a medium that selects for molecular linkage on the same DNA molecule of the third and the fourth marker sequences.

36. (Withdrawn) Plasmid pMXY9, comprising adjacently the URA3 marker gene and the CAN1 marker gene, whereby the two marker sequences flank a polylinker sequence for inserting a DNA sequence to be recombined and whereby the two markers are flanked by targeting sequences homologous to the BUD31-HCM1 locus on chromosome III of the *S. cerevisiae* genome.

37. (Withdrawn) Plasmid pMX9 according to claim 36, wherein the polylinker sequence comprises restriction sites for the restriction enzymes SmaI, XbaI, PacI and Bg/II.

38. (Withdrawn) Plasmid pMXY12, comprising adjacently the TRP1 marker gene and the CYH2 marker gene, whereby the two marker sequences flank a polylinker sequence for inserting a DNA sequence to be recombined and whereby the two markers are flanked by targeting sequences homologous to the BUD31-HCM1 locus on chromosome III of the *S. cerevisiae* genome.

39. (Withdrawn) Plasmid pMXY12 according to claim 38, wherein the polylinker sequence comprises restriction sites for the restriction enzymes SmaI, SpeI and PacI.

40. (Withdrawn) *S. cerevisiae* strain MXY47, characterized in that diploid cells thereof are homozygous for the alleles *ura3-1*, *trp1-1*, *can1-100* and *cyh2R* and heterozygous for the *msh2::KanMX* mutation.

41. (Withdrawn) *E. coli* strain JMI 01, containing plasmid pMXY9.

42. (Withdrawn) *E. coli* strain DH5, containing plasmid pMXY12.
43. (Withdrawn) Kit comprising at least a first container which comprises cells of *S. cerevisiae* strain MXY47, a second container which comprises cells of *E. coli* strain JM101 containing plasmid pMXY9 and a third container comprising cells of *E. coli* strain DH5.alpha. containing plasmid pMXY12.
44. (Withdrawn) Kit comprising at least a first container comprising cells of *S. cerevisiae* strain MXY47, a second container comprising DNA of plasmid pMXY9 and a third container comprising DNA of plasmid pMXY12.